THE EFFECTS OF SEASON, HOST PLANT PROTECTION, AND ANT PREDATORS ON THE SURVIVAL OF EUMAEUS ATALA (LYCAENIDAE) IN RE-ESTABLISHMENTS

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ABSTRACT. The primary purpose of this study on *Eumaeus atala*, the atala butterfly, was to determine which factors influence larval survival during re-establishment of atala butterfly populations. An inexpensive protective cover of fabric netting over the host plants at the re-establishment site was found to have a positive effect on the number of larvae that survived to pupation. Season was also found to have an effect on the number of re-established larvae that survived to pupation. Significantly more larvae survived to pupation during the wetter summer season than during the drier winter season. This suggests that future attempts to re-establish the atala should take place in the summer and should consider the use of protective netting over host plants. In the course of this study, the mortality of atala eggs was found to be high, and two new ant predators of atala eggs were found.

Additional key words: coontie, cycasin, insect.

The atala, *Eumaeus atala* Poey, is a hairstreak butterfly (Lycaenidae, subfamily Theclinae) with a tumultuous history. It was once considered to be extinct throughout south Florida due to the exploitation of its sole native larval host plant, but the atala has since made a comeback. There are close to 50 lycaenid species in the West Indies and south Florida (Smith et al. 1994). Female atala lay whitish-yellow eggs in clusters of up to thirty eggs which are usually deposited on fresh leaves of cycad species or on their reproductive cones. Atala larvae are a rusty-red color, with seven pairs of canary yellow spots on the dorsal side and short black setae. Their bright coloring is aposematic, as larvae and adults contain cycasin (Bowers & Larin 1989, Nash et al. 1992). Spiders, for example, who find the atala in their webs, avoid them (Hubbuch 1991). The atala apparently concentrate the secondary compounds of their larval host plant, the Florida coontie, Zamia pumila (Zamiaceae), in their bodies.

Eumaeus atala had a historical range of Dade, Monroe, and Broward counties in Florida. It also ranged throughout Cuba, and into the Bahamas (Clench 1943). In Florida, the atala's current range includes Dade, Broward, Monroe, Palm Beach, Indian River and St. Lucie counties (Culbert 1995). It also still occurs in Cuba, the Bahamas, and Cayman Brac (Hammer 1995).

Early scientific references to the atala describe the species as abundant in south Florida and Cuba (Scudder 1875, Schwartz 1888, Healy 1910, Grossbeck 1917). By the mid-twentieth century, the atala was considered rare or extinct in Florida by lepidopterists (Ford 1946, Klots 1951, Young 1956, Funk 1966, Rawson 1961). In 1979, on Key Biscayne, Florida, Miami-Dade county naturalist Roger Hammer found a thriving colony of *Eumaeus atala* (Roger Hammer pers. com.). Hammer attempted re-establishments, many of which were successful.

Atala multiplied at various locations in South Florida from Coral Gables to Florida City (Landolt 1984). "The atala has made a spectacular recovery and is now found in urban and natural areas around Fort Lauderdale and Miami" (Emmel & Minno 1993). The Key Biscayne colony, however, vanished for unknown reasons around 1991.

The main goal of this project was to conduct an atala re-establishment and to examine three questions. The first was to find out whether season would have an effect on larval and pupal mortality in a re-establishment. The second question was whether protecting atala host plants would increase survivability of larvae. It was hypothesized that if netting were placed over the host plants, more of the translocated larvae would survive compared to larvae on unprotected plants. The third question was are there predators of atala larvae, eggs and pupae, despite their cycasin content. This is important information because despite the fact that reestablishments appear to have preserved the atala in Florida, few details are known about what factors enhance success in re-establishment.

All of the study sites for this project were in Bear Cut Preserve, Crandon Park on Key Biscayne, Florida. Restoration of habitat was necessary prior to attempting atala re-establishment. The coastal maritime hammock where the study took place had been damaged by fire and exotic plant species (Doren et al. 1991). Few coontie plants remained in Crandon Park. Thirtyone coontie plants were purchased and planted in the re-establishment areas to improve the atala habitat prior to this project.

MATERIALS AND METHODS

Atala translocation to Key Biscayne began in January of 1999. Miami's Fairchild Tropical Garden donated all larvae and pupae used in the reestablishment. Fairchild was chosen as a source since it has the most consistently strong atala population in the county and because its colony originated from the

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1980's human-established atala colonies. The number of larvae used in releases for this project and the timing of releases themselves were always dependent on the population size of the Fairchild colony.

Atala larvae were all close to the same age at translocation, determined by measurement to be in their second instar. They were removed from the colony at Fairchild Tropical Gardens and placed on Z. *pumila* at four different sites on Key Biscayne with similar plant cover, topography, accessibility to host plants, and light levels. In addition to the larvae placed at these sites in Crandon, corresponding larvae were placed at an offsite location. Larvae were placed in three different treatment situations.

The three different treatments to the larvae were as follows. In Treatment #1, atala larvae were released and placed on Z. pumila in Crandon Park in a "natural" release with no treatment. Larvae had no protection from predators, except for their natural defenses. In Treatment #2, larvae were placed on Z. *pumila* in Crandon Park, then covered with a fine green nylon netting fabric that was tightly tied at the base of the plant. Holes in the mesh were 10 mm. Upon pupation of the atala, this netting was removed. In Treatment #3, the larvae were reared in captivity off-site on freshcut Z. pumila fronds under a high level of protection on a screened porch without temperature control. This third treatment gives an indication of how survival of atala in captive rearing compares to survival in the field.

The attempted re-establishments in Crandon Park and the off-site captive rearing experiments were repeated three times throughout the year: Winter/Dry season, Transitional/Spring season and Summer/Rainy season (see Table 1).

Length of residency was used as a measure of survival for larvae. If a larva was no longer on the host plant or was not discovered pupating on other plants within 1 m of the host plants, it was considered to have died, even if the actual "corpse" was not discovered. Although larvae sometimes leave the host plant to pupate, they tend to do so in clusters and are easily found usually within 1 m of the host plant regardless of whether netting is present. This was reinforced in my experiment at an off-site location on a screened porch where, despite the lack of netting around coontie, 76% of atala larvae did not venture farther than 0.5 m from their point of translocation to pupate. The other 24% of larvae crawled up to 1 m away, but never to a greater distance when sufficient coontie was available, despite there being no barriers to prevent them from doing so. At both the park and the offsite location, while there were other plants surrounding the translo-

Date	Experiment	Total # of larvae	Treatment	Sites released
1/15/99	#1	59	none	1, 2, 3, 4, 5
1/28/99	#1	59	captivity	ex situ
2/16/99	#1	59	netting	1, 2, 3, 4, 5
4/9/99	#2	21	none	2
5/5/99	#2	21	captivity	ex situ
5/5/99	#2	21	netting	2
6/30/99	#3	54	none	1, 2, 5
6/30/99	#3	54	captivity	ex situ
6/30/99	#3	54	netting	1,2,5

cation sites, the only coontie available were those that were part of the experiments.

Pupal mortality was distinguished by presence and condition of the pupae. When an atala butterfly emerges naturally, there is a cleanly consistent ecdystal slit in the cuticle. If the pupa has been opened by outside force, this is easy to discern. If the pupa vanished, it was assumed to have been taken by a predator unless discovered in its entirety on the ground beneath the plant on which it pupated.

Ideally, Treatment #1, Treatment #2, and Treatment #3 would all have been initiated simultaneously at the beginning of each of the three seasons. However, larvae were not always available in sufficient numbers at Fairchild, so some of the differing larval treatments in the same experiment were separated by 2–4 week time spans (still within the same season).

The four sites in Crandon Park, sites #1, 2, 3, and 5 were assumed to be similar to one another in plant cover, light level and topography and were used as replicates (Site 4 was removed from the study prior to its onset). Sites 6–13 were off-site captive-rearing sites assumed to be similar to one another and used as replicates.

To address the hypothesis that netting improved the ability of larvae to survive to pupate and the question of whether season impacted larval survival, analyses of variance were performed by treatment and season. Pupal survival to emergence was also examined in these ANOVAs. Tukey's post hoc test was also performed.

RESULTS

There was no interaction between treatment and season on larval survival to pupation, F (4, 15) = 2.30, p = 0.107. There was also no interaction between treatment and season on survival to emergence, F (4, 15) = 0.35, p = 0.842.

TABLE 2. Main effects of season and treatment on atala pupation and emergence.

Analyses of variance Main effects of	df	F	Significance
TREATMENT on % pupating	(2, 15)	16.13	p < 0.001
TREATMENT on % emerging	(2, 15)	3.08	p < 0.076
SEASON on % pupating	(2, 15)	6.86	p < 0.008

There was a main effect of treatment on pupation, F (2, 15) = 16.13, p < 0.001 (Table 2). Tukey's post hoc test at a 5% significance level demonstrated that the mean percentage of atala pupating with protective netting (62.7%) was significantly greater than the mean percentage pupating without netting (17.1%) (Table 3).

There was also a marginally significant main effect of treatment on emergence, F (2, 15) = 3.08, p < 0.076 (Table 2). Tukey's post hoc test at a 5% significance level revealed that the mean percentage of atala that emerged from their pupae successfully on plants where netting had been applied earlier on (96.9%) was significantly higher than the mean percentage that emerged from their pupae without netting ever having been used (60.8%) (Table 3).

As predicted, there was a significant main effect of season, F (2, 15) = 6.86, p < 0.008 (Table 2). Tukey's post hoc test indicated that Season #3/Summer had a significantly higher percentage (56.8%) of pupation than Season #1/Winter (33.2%) (Table 3).

Few predators of the atala in any of its life stages have ever been reported. In the course of this study, several ant species were found to be major predators on atala in the egg stage, and one species was found to enter pupae. The first ant species noted as an egg predator was *Camponotus abdominalis* var. *floridana*. This is a common native ant in Florida that often infests dwellings. Also called "bull dog" ants, they are known to feed on honeydew and insects (Smith 1972). In prior lab feeding trials, atala adults and cycasin were considered deterrents to *C. abdominalis* (Bowers & Larin 1989).

The same species of ant was observed at Site #2 in Crandon on 4 June 1999 tearing apart atala eggs where 19 eggs had been laid. At the same time, an atala butterfly was fluttering around the plant in a pattern typical of an egg-laying female. The butterfly got very close to the frond where the ant was eating eggs, and beat at the ant with its wings. The ant grabbed an antenna of the atala butterfly and pulled. There was a fierce struggle, and the butterfly fell to the ground. The atala beat its wings on the ground, momentarily unable to fly, then flew away quickly. This unusual behavior appeared to be very purposeful on the part of the atala, as if it were attempting to drive the ant away from its offspring. TABLE 3. Tukey's post hoc test at 5% level: mean percentage of atala pupation and emergence by treatment group and by season.

	Mean % pupating:	Mean % emerging:
BY TREATMENT GROUP		
#1 Group (with No Treatment) ^a	17.1%	60.8%
#2 Group (with Medium	62.7%	96.9%
Treatment/Protection Level) ^b		
#3 Group (with High Protection)	° 54.7%	96.7%
Significant difference:		
Group #1 ^a vs. #2 ^b	p < 0.001	p < 0.046
Group $#2_{\rm b}$ vs. $#3^{\rm c}$	n.s.	n.s.
Group #1 ^a vs. #3 ^c	p < 0.001	p < 0.047
BY SEASON	-	ŕ
#1 Scason ^d	33.2%	82.3%
#2 Season ^e	55.6%	84.6%
#3 Scason ^f	56.8%	88.2%
Significant difference:		
Season #1 ^d vs. #3 ^f	p < 0.009	n.s.
Season #1 ^d vs. #2 ^e	n.s.	n.s.
Season $#3^{f}$ vs. $#2^{e}$	n.s.	n.s.

* In situ without netting on plants

^b In situ with netting on plants

[°] Ex situ in captivity (outside cages)

^dWinter/Dry Season ^eTransitional/Spring Season

^fSummer/Rainy Season

n.s. = no significance, p > 0.05.

It took a *Camponotus* ant very little time to find atala eggs. On 30 August 1999, a single ant was observed at Site #2 at 13:32 on a coontie frond while an atala was laying eggs. The ant came near enough to the six eggs to attack them, but did not, then turned around and left the plant. Ten minutes later, another ant (possibly the same one) appeared and began to tear a hole in the eggs. Within eighteen minutes of the egg-laying event, three other ants of the same species arrived and began a new attack on the eggs, until all six were destroyed.

On 5 July 1999 a large ant (*Pseudomyrmex mexi*canus) was observed attacking atala eggs at Site #2 where 25 eggs had been laid. Nine undisturbed pupae were also present on the same plant. *Pseudomyrmex mexicanus* is native and found throughout the eastern United States (Smith 1972).

Another egg predator was observed during a survey at Rockdale Pineland in Miami on 3 October 1999. *Wasmannia auropunctata* (Roger) spent 15 min puncturing a small hole in an egg before the ant was collected. Commonly known as the "little fire ant," *W. auropunctata* is a neotropical ant introduced into Florida (Smith 1972).

For several weeks, pupae would occasionally be found in Crandon Park with a perfectly round 0.1 cm diameter hole in the outer layer. Several times, upon examination, tiny ants (*Monomorium floricola*) spilled



FIG. 1. A female *Eumaeus atala* (atala butterfly) depositing eggs on a *Zamia pumila* (coontie) frond. Photograph by Robert Schroeder (used with permission).

out of the holes. *Monomorium floricola* is an introduced ant from Africa or Asia, known to feed on insects (Smith 1972). These ants may have been opportunists who went into holes made by a parasite.

In Bear Cut Preserve, Crandon Park, ants appeared to be a major cause of egg mortality and had a serious effect on atala survival. Ants left visual evidence behind in the form of characteristic broken and torn apart eggshells. Ants were also observed, less commonly, carrying eggs away from the plant. Over 700 atala eggs were observed to have been laid in Crandon Park. Of these, 131 were destroyed in a manner that implicated ants and at least the same number of eggs simply vanished.

DISCUSSION

The results of this re-establishment provide input for a plan of successful re-establishment of the atala. The survival of translocated larvae was greatly enhanced by a simple and inexpensive protective netting treatment that affords extra protection in the vulnerable larval stage. Survival of these protected in situ larvae was equivalent to the survival of highly protected captive-reared larvae. It is therefore recommended that atala re-establishments use protective netting to cover larvae and plants until at least pupation. Reestablishment utilizing pupae rather than larvae should also be tested, since once atala pupate, survival is quite high, even without netting protection.

As soon as all larvae in the experiments had pupated, the netting was removed, so the use of netting might not necessarily be expected to affect the percentage of atala that survived from pupation to emergence. Despite this, there was marginal significance, with more butterflies successfully emerging from pupae that had been protected by netting in their larval stage. Further testing should be conducted to determine how the netting utilized in the larval stage may enhance future pupal survival.

Larval survival was highest in the summer. The summer season is also the wettest. It is a logical outcome since during the rainy season, there are more fresh coontie fronds available, which are the favored food of early instars. During the dry season, the young atala caterpillars must work harder to survive, scraping at the underside of old, tougher fronds. Reestablishments of the atala appear to have a much greater chance of success when conducted in the summer rainy season.

Egg protection must be factored into the re-

establishment equation. The impact of native and exotic ant species on the atala requires a detailed examination. Finer netting, placed over eggs as soon as possible, may discourage these and other predators.

Re-establishment of the atala into habitat where it was once common, but has been locally extirpated, is possible with a limited time commitment and limited financial investment, and the results appear to be very promising. The re-established atala colony on Key Biscayne, with no translocations having been done since June of 1999, was still abundant at the time of the last survey by this researcher in November of 2001.

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